

The results of table 6 show that the detection level or the binding of C. atrox venom was 3.7 µg in normal rabbit serum but dropped to 1.2 µg in presence of homologous rabbit anti C. atrox serum. Thus, showing the difference of 2.5, which is the neutralizing index for this antiserum. The venom neutralized by the specific anti venom is not detected by anti-LTNF. The neutralizing index depends upon the potency of the anti serum. The neutralizing index for anti serum to V. russelli was 2.9 and for O. scutellatus 0.6

### CLAIMS

#### **We claim:**

- 15 1. A composition of matter comprising Anti-LTNF, which is polyclonal or monoclonal antibody made versus:

natural LTNF-n having mol. wt. of 68 kilodalton and a partial amino acid sequence from the N-terminal of fifteen amino acids

Leu Lys Ala Met Asp Pro Thr Pro Pro Leu Trp Ile Lys Thr Glu.

- 20 2. The composition of matter comprising Anti-LTNF including polyclonal or monoclonal antibodies made versus

any active portion of LTNF-n sequence and specifically those antibodies to LTNF-15, LTNF-10 and LTNF-5, comprising of 15, 10 and 5 amino acids, respectively, from the N-terminal of LTNF-n.

3. A composition of matter as in claim 2, wherein the polyclonal anti-LTNF is further characterized as immunoglobulin (IgG) from immunized animals and monoclonal anti-LTNF in claim 2 is further characterized as immunoglobulin (IgG) from fused hybridoma cells from immunized mouse or mouse myeloma cells, or other appropriate cells.

4. The in vitro toxin assay process, based on the use of Anti-LTNF (made versus LTNF-n, LTNF-15, LTNF-10 or LTNF-5) as a reagent for the detection of biological toxins, becomes an ethical replacement for

currently used animal bioassay, typically mice, or other rodents and other animals. *B/*

*Sub B2* 5  
5. The anti-LTNFs made versus natural LTNF and versus synthetic peptides consisting of at least five amino acids detect biological toxins derived from animal, plant and bacteria by ELISA.

10 6. The anti-LTNFs made versus LTNF-n and the synthetic peptides comprising of 15, 10 and 5 amino acids recognize toxins derived from animal, plant and bacteria by ELISA assay. *B/*

15 7. The anti-LTNFs provide essential reagents for the in vitro assay of the wholesomeness of toxins existing in singular form, or in mixture, in a manner comparable to animal bioassay.

8. The anti-LTNFs detect and assay the toxins from foods, blood sera and other body fluids saliva, milk, urine etc. by ELISA test in antigen capture format, or any similar test.

*Sub B3* 20  
9. The neutralizing potency of an anti-toxin is the neutralizing index given by the toxin assay minus an anti-toxin mixture assay; wherein, the toxin assay is determined by ELISA test of the toxin plus normal serum; and the anti-toxin mixture assay is determined by ELISA test of a mixture of toxin plus anti-toxin mixture, such mixture containing a reduced amount of free toxin due to neutralization by the anti-toxin.  
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*a* 30  
10. The neutralizing potency of anti-toxins including anti-venoms can be assayed by in vitro test using anti-LTNF compositions as in *claim 2* and 2 as reagent, and thus saving thousands of mice as well as time and money.

35 11. A composition of matter comprising an antibody made versus a peptide containing at least five amino acids from the N-terminal sequence

Leu Lys Ala Met Asp Pro Thr Pro Pro Leu Trp Ile Lys Thr Glu.

12. A composition of matter as in claim 11, which is in the form of an immunoglobulin selected from the group consisting of an immunized animal serum, a hybridoma cell culture and a mouse ascitic fluid.

13. A composition of matter as in claim 12, which reacts immunologically with a toxin selected from the group consisting of an animal toxin, a plant toxin and bacterial toxin.

14. A process comprising contacting, in vitro, a biological toxin with an antibody made versus a sequence of at least five amino acids from the N-terminal of the sequence

Leu Lys Ala Met Asp Pro Thr Pro Pro Leu Trp Ile Lys Thr Glu

under conditions to cause the biological toxin to react immunologically with said antibody.

15. A process as in claim 14, wherein the novel antibody is made against LTNF having a non-immunological binding with toxins such that its antibody has the property of being able to react immunologically in vitro with a wide range of biological toxins.

16. ~~Processes~~ <sup>A Process</sup> as in claim 15 which is carried out according to an ELISA double-sandwich method protocol.

17. A process as in claim 15, wherein the biological toxin reacts immunologically with said novel antibody to produce a first reaction product, said process further comprising contacting the first reaction product with a set of antibodies made for specific known toxins for the purpose of producing a modified ELISA test capable of identifying toxins with the help of the novel anti-LTNF.